HIGH SENSITIVE DIOXIN ANALYSIS FOR HUMAN BLOOD BY 7ML SAMPLE SIZE

Matsumura T.⁽¹⁾, Morita M.⁽²⁾, Miyata H.⁽³⁾, Suzuki N.⁽⁴⁾, Watanabe M.⁽⁵⁾, Ishiguro Y.⁽⁶⁾ and Iida T.⁽⁷⁾

(1) Environmental Risk Research Center, Institute of General Science for Environment, IDEA Consultants Inc., 1334-5 Riemon, Ohigawa, Shida, Shizuoka 421-0212, JAPAN, (2) Ehime university, (3) Faculty of Pharmaceutical Sciences, Setsunan University, (4) National Institute for Environmental Studies, (5) National Institute of Health and Nutrition, (6) Fukuoka Institute of Health and Environmental Sciences, (7) Kitakyushu Life Science Center

Abstract

High sensitive detection method for dioxins in human blood using SCLV Injection System was developed. This method is possible to reduce sample size for dioxin analysis of human blood to 7mL at a level of low pg-TEQ/fat-g detections.

Introduction

The authors has been developed SCLV (Solvent Cut Large Volume) Injection System for the trace analysis of PCDDs, PCDFs and PCBs at levels as low as a few femto (10^{-15}) grams per microlitre. This system is possible to reduce sample size for dioxin analysis of human blood. Usually, analysis of human blood at a level of <10pg-TEQ/fat-g requires 40-60mL sample size (ex. final solvent volume: 10uL, fat % of blood: 0.3-0.7%, GC/MS sensitivity: S/N>5-10 at 10fg). Blood-gathering at 40-60mL size using "blood-gathering bag" is sometimes overburdened with mental anguish / bodily pain for test subjects, especially for infants/aged persons. Using this SCLV Injection technique, 7mL sample volume is enough to evaluate for dioxin concentration in human blood with accuracy. 7mL blood-gathering make possible to use vacuum test tube as blood-collecting equipment. Sampling became as undergo like a medical health examination/inspection.

Methods and Materials

Schematic of SCLV Injection System use for this study is shown in *Figure-1*. Detection of PCDDs, PCDFs and dioxin-like PCBs were carried out by HRGC/HRMS method after liquid/liquid extraction and gel clean-up procedures. Analytical procedure is represented in *Figure-2*. Concentration of PCDDs, PCDFs and dioxin-like PCBs were determined by HRGC (6890, Agilent Technologies)/HRMS (AutoSpec-Ultima, Micromass) equipped by SCLV Injection System (SGE Japan Inc., Japan). Twenty-nine (WHO-1998's) native (Wellington Laboratories, Canada) and ¹³C- isomers (Wellington Laboratories, Canada) were used as calibration standards and internal spikes. To detect low pg/L concentrations of PCDDs, PCDFs and PCBs, organic solvents used for

analysis were purified by sub-boiling distillations. Glassware and silica gel use for pre-treatment were cleaned by heating at temperature of 450 and 400 after an organic solvent wash. All process was carried out in a chemical hazard clean room (ca. class<10000). BPX-Dioxin-I (ID: 0.15mm, length: 30m, SGE) are equipped for HRGC using SCLV Injection System. 7mL blood sample were collected by 7mL glass vacuum testing tube (VACUTAINER^{TR} (7mL, 13 x 100mm, Sodium Heparin 100 U.S.P. Unit), Becton, Dickinson and Company). Analyses were performed in pursuance of ISO/IEC 17025 (JCLA4, Japan Chemical Laboratory Accreditation).

Results

As an example of results, chromatograms for tetra-octa CDDs and CDFs are shown in *Figure-3* together with assignation of 17 kinds of 2,3,7,8-substituted isomers and fat base concentration of each isomer. Detection limits calculated by S/N ratio were 0.1-1.0 pg/g-fat for each isomer (depends on fat % and type of isomers). These results represent this method is available for dioxin analysis for human blood at low pg-fat concentration level at low sample size (7mL).

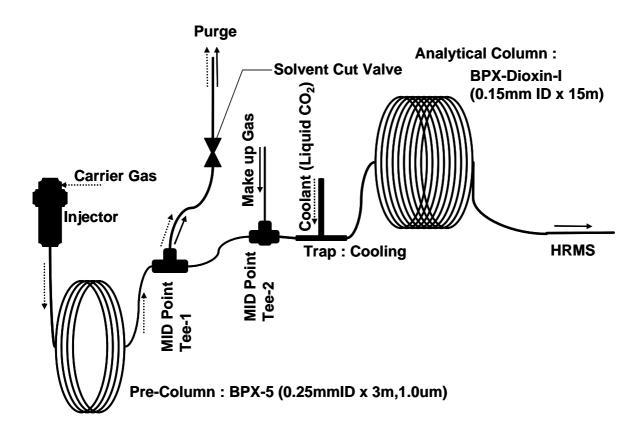


Figure-1. Schematic of SCLV Injection System use for this study.

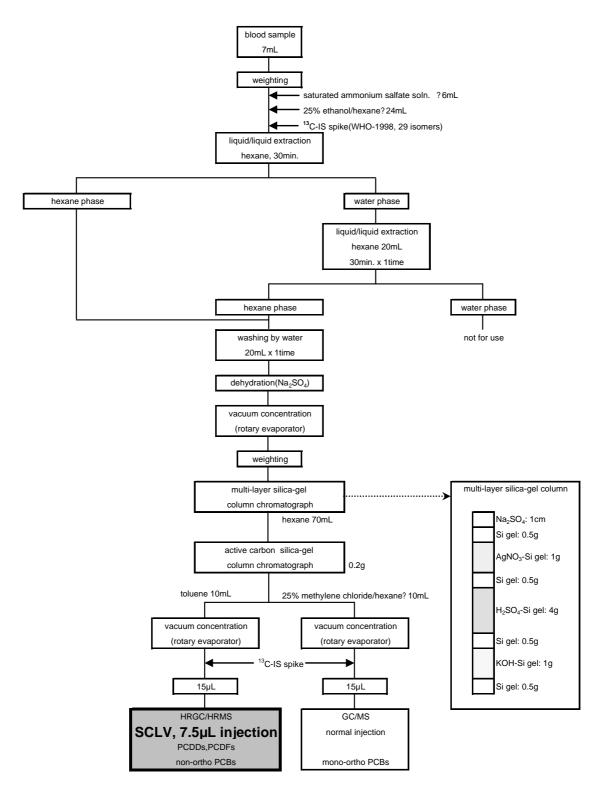


Figure-2. Analytical procedure for PCDDs, PCDFs and dioxin-like PCBs for this study.

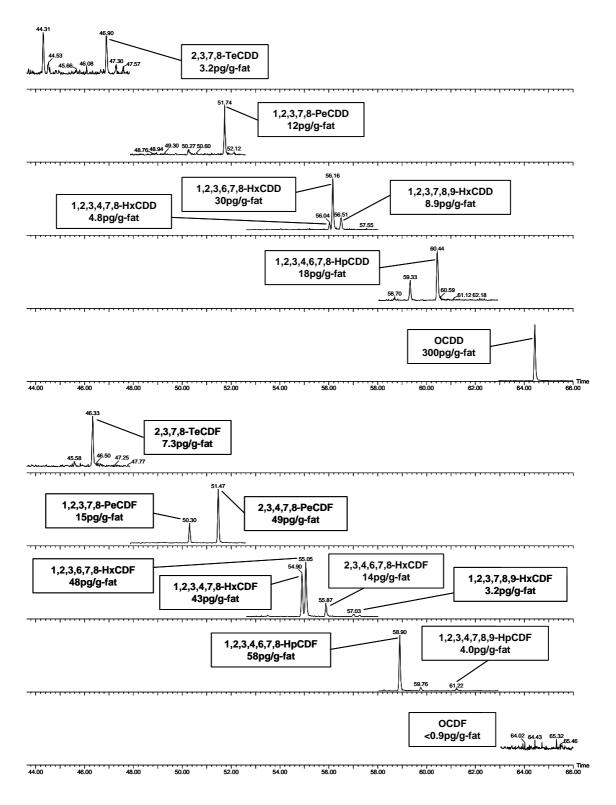


Figure-3. Example of chromatogram of tetra, penta, hexa, hepta and octa CDD(s) and CDF(s).